

Far Eastern Entomologist

Дальневосточный энтомолог

Journal published by
Far East Branch of the
Russian Entomological Society
and Laboratory of Entomology,
Institute of Biology and Pedology,
Vladivostok

Number 43: 1-8

ISSN 1026-051X

April 1997

MORPHO-KARYOLOGICAL DESCRIPTION OF *PAGASTIA ALTAICA* SP. N. (DIPTERA, CHIRONOMIDAE) FROM ALTAI MOUNTAINS, WITH THE KEY TO HOLARCTIC SPECIES OF *PAGASTIA* OLIVER

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The description of male and karyotype of fourth instar larva of *Pagastia altaica* sp. n. (subfam. Diamesinae) from Katun' River (Altai Mountains) are given. New species resembles *P. orientalis* (Tshern.), but differs by structure of male hypopygium and by karyotype of larva, which consists of the four pairs of chromosomes (one metacentric, two submetacentrics and one acrocentric). Three long polytene and a "pompon"-like chromosomes are detected in the salivary gland cells. A series of chromosome markers are described.

KEY WORDS: Diptera, Chironomidae, *Pagastia*, new species, karyotype.

Е.А. Макарченко¹⁾, И.Е. Керкис²⁾, О.В. Иванченко²⁾. Морфо-кариологическое описание *Pagastia altaica* sp. n. (Diptera, Chironomidae) с Горного Алтая, с определительной таблицей видов *Pagastia* Голарктики // Дальневосточный энтомолог. 1997. N 43. С. 1-8.

Приведено описание самца и кариотипа личинки IV возраста нового вида хирономид подсем. Diamesinae *Pagastia altaica* sp. n. из реки Катунь (Горный Алтай). Новый вид на всех стадиях развития очень близок к *P.*

orientalis (Tshern.), от которого отличается строением гипопигия самца, структурой помпоновидной хромосомы и расположением колец Бальбиани в политечных хромосомах слюнных желез личинки.

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INTRODUCTION

Genus *Pagastia* Oliver, 1959 was described from North America and before our investigations included only four Nearctic species: *P. orthogonia* Oliver, 1959, *P. partica* (Roback, 1957), *P. sequax* (Garrett, 1925) and *Pagastia* sp. A. (Oliver, 1959, 1983, 1986, 1989; Oliver & Roussel, 1982). Later, Makarchenko (1981, 1985, 1989, 1994) revised some *Syndiamesa* Kieffer, 1918 and *Pseudodiamesa* Goetghebuer, 1939 from East Palaearctic and removed *S. orientalis* Tshernovskij, 1949, *S. angarensis* Linevich, 1953 and *S. lanceolata* Tokunaga, 1936 to *Pagastia*.

Adult male, pupa and larva of *Pagastia* are similar to that of *Pseudodiamesa*. Primary *Pseudodiamesa*-group included genera *Pseudodiamesa*, *Pagastia* and *Hesperodiamesa* Sublette, 1967. Their systematics was very entangled. Firstly we compare morphology of males, pupae, and larvae of *Pseudodiamesa* and *Pagastia* and made a conclusion that *Pagastia* is subgenus of *Pseudodiamesa* (Makarchenko, 1989). *Pseudodiamesa* o. *orientalis* (Siberia and Russian Far East mainland) and *P. orientalis* *insularis* Makarchenko (Sakhalin and Kuril Islands) have been recorded also (Makarchenko, 1989). Later, *P. o. insularis* was mistakenly synonymized with Japanese *P. lanceolata* (Makarchenko, 1993, 1994). It was happened because of the holotype of *P. lanceolata* described by Tokunaga (1936) is lost. Karyological studies confirmed that *P. orientalis* and *P. lanceolata* are related separate species and *Pseudodiamesa* and *Pagastia* are separate genera (Kerkis, 1992). However, study of the systematics of genus *Pagastia* is not completed yet and more detail information including new data on synonymy of known species will be given in separate paper.

During investigation of male morphological characters and larval polytene chromosomes of *Pagastia* from Altai Mountains we discovered that *P. orientalis* and *P. altaica* sp. n. are distributed here. The holotype of new species is deposited in Institute of Biology and Pedology (Vladivostok).

MATERIALS AND METHODS

Specimens of *P. altaica* sp. n. were fixed in either Udemans solution (adults) or 70% ethanol (pupae and larvae), treated in 10% KOH solution for maceration of soft tissues, and finally mounted for microscopy in Euparal and Fora-Berleze solutions.

Fourth instar larvae were used in karyological analysis. Age determination based on the size of imaginal discs in the first thoracic segment (Wülfel &

Gots, 1968). The larvae were fixed in a 3:1 absolute ethanol-acetic acid and stored in refrigerator. Analysis of the polytene chromosomes was performed on squashed salivary glands prepared by the standard aceto-orcein technique. Chromosome arms were designated by letters from A to G, and each arm, of polytene chromosome was marked in the direction from the telomere to the centromere according to Keyl's karyotaxonomics for Chironominae (Keyl, 1962) particularly in its appliance to the *Chironomus* genus. It should be noted, however, that there were no homologies between the banding patterns of the species under study and the species of the genus *Chironomus*.

In analysis of chromosomal polymorphism we used the designation system accepted for the Chironominae subfamily (Kiknadze et al., 1991). Thus to identify the active regions staining with methylene green with pyronine was used. The karyotype of *P. orientalis* was taken as standard in the descriptions of the karyotype of the *P. altaica* sp. n.

Abbreviations and terminology in the description of the adult, follow Saether (1980). For some structures of hypopygium, namely for gonocoxal lobes and aedeagal lobes, terms of Hansen & Cook (1976) were used.

DESCRIPTION OF NEW SPECIES

***Pagastia altaica* Makarchenko, Kerkis et Ivanchenko, sp. n.**

(Figs 1, 2)

MATERIAL. Holotype: ♂, Katun' River, about 3-5 km from Chemal Vill, Altai Mnts., Russia, 29.IV 1989 (leg. E. Makarchenko). Paratypes: 2♂♂, 1 pupa, 3 larvae (70% ethanol), 3 larvae (Karnua, 3:1), the same data as holotype.

DESCRIPTION. MALE imago (n=3). Generally colour from dark brown to black, wing greyish. Body length 5.0-5.2 mm. Wing length 4.7-4.8 mm.

Head. Coronals 4 (22.4-32.0 µm), outer and inner verticals about 12, orbitals 14, postorbitalis 24, clypeals 30-34. Subapical seta of terminal segment antenna flagellomere length 32-61 µm. AR=2.7-2.9. Last 4 maxillary palp segments length (µm): 131-176 : 243-330 : 266-342 : 330-403. Sensilla capitata diameter 6.0 µm. Head width/palp length 0.7-0.8.

Thorax. Antepronotum with 9-12 (65-100 µm) median and 16-20 (110-190 µm) ventrolateral setae. Acrostichals 12-17 (60-70 µm), dorsocentrals 22-23, prealars 22-32, scutellars about 40.

Wing. Top of wing without macrotrichiae. R and R_1 with 25-27 (35-38 µm) macrotrichia, R_{4+5} with 8-12 macrotrichia. Squama dark grey, with 57 (110-220 µm) setae.

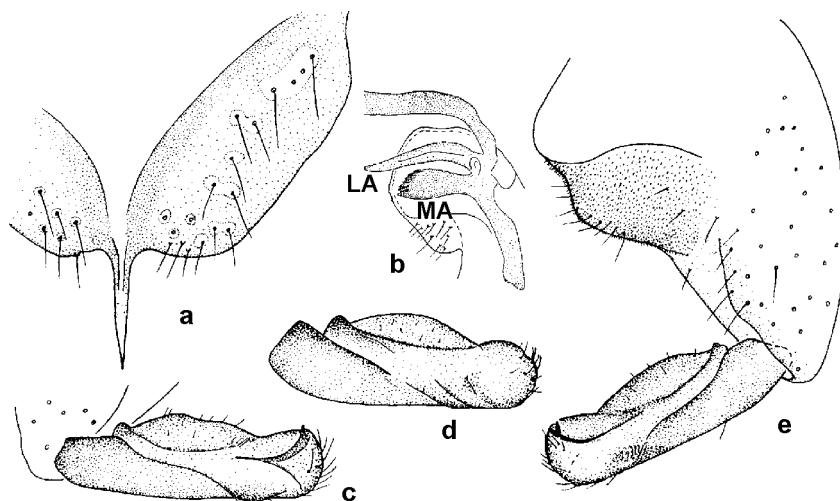


Fig.1. Male hypopygium of *Pagastia altaica* sp. n., holotype: a) tergite IX with anal point; b) aedeagal lobes; c, d) gonostylus; e) gonocoxite and gonostylus. LA - lateral aedeagal lobe, MA - median aedeagal lobe.

Legs. Front and hind legs with long setae, $BR_1=7.2$, $BR_2=3.8$, $BR_3=4.6$. Front tibial spur length 169.6 μm , middle tibial spurs length 131.2 μm and 128 μm , hind tibial spurs length 121.6 μm and 169.6 μm and with comb from 14 (67.2-115.2 μm) spines. Length (μm) and proportions of legs:

P	f	t	ta ₁	ta ₂	ta ₃	ta ₄	ta ₅	LR	SV	BV
P ₁	2252	2669	1960	1126	709	417	328	0.73	2.51	2.67
P ₂	2335	2585	1376	792	500	292	292	0.53	3.58	3.36
P ₃	2627	3169	1918	1001	584	375	328	0.61	3.02	3.37

Hypopygium (Fig. 1). Anal point narrow, without apical peg, 105.6-108.8 μm length. Phallapodeme with median and lateral aedeagal lobes. Median aedeagal lobe length 392-411 μm , apex digitated; lateral aedeagal lobe length 256-270 μm . Tergite IX with 30-38 (35-140 μm) setae, laterosternite IX with 11-20 setae. Basal plate and lobe-like median field of gonocoxite with numerous setae. Gonostylus slightly costated, subapical rounded, without "heel", megaseta length 12.8-25.6 μm . Gonostylus length/gonostylus width 2.6-2.9. HR=1.3.

PUPA and LARVA are not distinguished by morphological features from those of *P. orientalis*.

KARYOTYPE OF FOURTH INSTAR LARVA. Chromosome number in *P. altaica* sp. n. is $2n=8$. Four chromosome pairs are seen on metaphase spreads of mitotically dividing cells. The chromosomes of one pair are

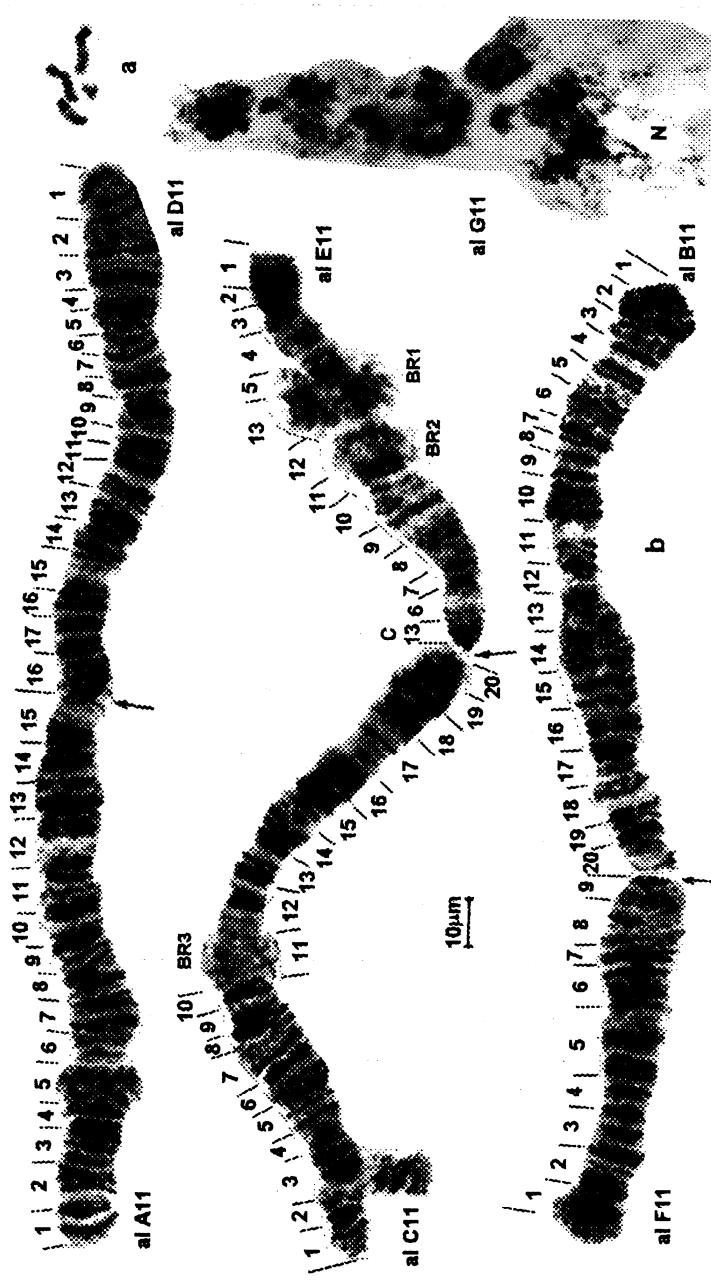


Fig. 2. *Pagastia altaica* sp. n., entire chromosome component: a - mitotic chromosomes, b - standard photomap of polytene chromosomes. A-F - arms of the chromosomes; G - "pompon"-like chromosome; N - the nucleous organizer; BR - Balbiani ring.

metacentric, they are submetacentrics in two pairs and acrocentric in the last pair (Fig. 2a). Three long polytene and a “pompon”—like chromosomes are detected in the salivary gland cells (Fig. 2b). The precentromeric regions of the chromosomes are united into a common pseudochromocenter presumably through ectopic contacts. The “pompon”—like chromosome is not involved in the formation of the pseudochromocenter. The centromeric regions of the polytene chromosomes are indistinct. The homologues tightly conjugate along almost all their lengths; short asynaptic stretches are, however, observed in some regions. Band sequence in arms A,B,C,D,F in *P. altaica* sp. n. is the same as in *P. orientalis*. Balbiani ring in region 11 appears in arm C in *P. altaica* sp. n. In contrast, it does not appear in *P. orientalis* and *P. lanceolata*. Arm E differ from standard or E11 by a single heterozygous inversion. These inversion was found in heterozygous condition in a single larva.

or E1 1 2 3 4 5 6 7 8 9 10 11 12 13
al E1 1 2 3 4 5 13 12 11 10 9 8 7 6 13

The “pompon”—like chromosome of *P. altaica* sp. n. appears as heterochromatin clumps with orderly arrangement. This is the feature making the “pompon”—like chromosome of *P. altaica* sp. n. morphologically different from that of *P. orientalis* and *P. lanceolata*.

Based on the results of karyological analysis, we made a conclusion that the species of the *Pagastia* are closely related. Males can be separate by features included in key below.

BIOLOGY. Larvae and pupae of new species inhabit big-sized stones and rocks of upper and middle parts of Katun' River. They build loose sand cases on upper and lateral surfaces of stones. These cases are similar to that of *P. orientalis*, like pancake and consist from grain of sand and many diatoms algae. Water temperature during the collecting was 8.2–9.5 °C.

DISTRIBUTION. Russia: Altai Mountains, Katun' River.

KEY TO MALES OF Holarctic *Pagastia* Oliver, 1959

1. Eyes hairy. Aedeagal lobes reduced. (Subgenus *Hesperodiamesa* Sublette, 1967).
Basal lobe of honocoxite is very small. Canada (British Columbia, Alberta)
..... *P. (H.) sequax* (Garrett, 1925)
- Eyes pubescent. One or two aedeagal lobes present. (Subgenus *Pagastia* Oliver, 1959) 2
 2. Median and lateral aedeagal lobes present. AR 2.5-4.2 3
 - Lateral aedeagal lobe presents only. AR 1.7-2.1 6
 3. Median aedeagal lobe digitated and widest in distal part 4
 - Median aedeagal lobe widest in middle and sharp-clawed apically. Canada (Yukon Territory), USA (Alaska, Utah, Wyoming, North Dakota, Washington) *P. (P.) partica* (Roback, 1957)
 4. Gonostylus subapical with “heel”. Russia (Sakhalin, South and Middle Kuril Islands), Japan (Hokkaido, Honshu) *P. (P.) nivis* (Tokunaga, 1936)

- Gonostylus subapical without “heel” 5
- 5. Gonostylus subapical angled. Russia (from Altai Mountains to continental part of Far East), China (Liaoning, Jilin, Heilongjing), South Korea
..... *P. (P.) orientalis* (Tshernovskij, 1949)
- Gonostylus subapical rounded (Fig.1). Russia (Altai Mountains)
..... *P. (P.) altaica* sp. n.
- 6. Anal point widest in basal part and thin in apical, pointed and often with peg. Russia (Baikal Lake basin, Primorye, Sakhalin, Kunashir Island), South Korea, Japan (Hokkaido, Honshu)
..... *P. (P.) lanceolata* (Tokunaga, 1936)
- Anal point finger-shaped, rounded apically and without peg. USA (Alaska, Michigan, North Dakota) *P. (P.) orthogonia* Oliver, 1959

ACKNOWLEDGEMENTS

The authors are grateful to Drs. A.G. Istomina and A.Yu. Kerkis (Institute of Cytology and Genetics, Novosibirsk, Russia) for help during the collection of the material in Altai Mountains. We are much indebted to Dr. H. Niitsuma (Shizuoka University, Japan) for sending the specimens of *P. lanceolata*. We also appreciate to Mrs A. Fadeeva for translation of the karyological part of manuscript in English.

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